On Archerina, Golenkinia and Botryococcus.

By

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With Plate 25.

A. Archerina Identical with Golenkinia and Richteriella.

In the year 1885 I described in this Journal—vol. xxv, new ser., p. 61—and figured in a coloured plate, a minute chlorophyllogenous organism, for which I formed the genus "Archerina." I named the species "A. boltoni." Nine years later [1] Professor Chodat, of Geneva, described the same organism under the name "Golenkinia radiata," in Morot's 'Journal de Botanique,' Tome viii, 1894, September 16th. Professor Chodat obtained his specimens from a small duck-pond in the public park at Geneva. My specimens were sent to me in a bottle-full of living material by Mr. Thomas Bolton, of Birmingham. I have now reason to believe that the "gathering" was obtained by Mr. Bolton from the duck-pond of the gardens of the Royal Botanical Society in Regent's Park, whence I have since obtained Archerina.

No one who compares my figures of the actinophrys-like form of Archerina (figs. 2, 4, 5, 6, 7, 13, 16, and 17 of Pl. 7, vol. xxv, 'Quart. Journ. Micr. Sci.,' New Series) with Chodat's figures and description can doubt that the organism figured by me is the same as that represented nine years later

[1] I am indebted for my knowledge of Professor Chodat's memoirs to a most valuable and compact little volume on the 'British Freshwater Algæ,' by Mr. G. S. West, published in the Cambridge Biological Series, 1894.

by Chodat (compare especially fig. 2 of his Pl. III). Not only are the radiating processes and the form of the chlorophyll bodies identical in the two sets of figures, but Chodat also figures and describes the empty, discarded spherical cases with radiating processes, which I described under the name of "ghosts," and figured in fig. 18 of my Pl. 7.

In examining this organism in 1884 I was particularly struck by the frequent association with it of colourless, naked amœboid protoplasm, which I was led—I now think erroneously—to consider as an essential part of the organism itself. I now believe that this amœboid protoplasm belonged to a Vampyrella-like organism which associated itself with the Archerina, and frequently invested it so closely as to lead to the supposition that it was part of the Archerina itself. I have since come across several cases of this close investment of a minute algoid organism by the naked protoplasm of an amœba-like or Vampyrella-like companion. A case which I may mention is that of the hollow botryoidal fronds of the interesting Clathrocystis æruginosa of Henfrey, which I have had very ample opportunity of studying.

I have no doubt that it is due to the fact that I was led, by the association of extraneous amœboid protoplasm with many specimens of Archerina, to refer this organism to the Protozoa, that my description of it has escaped the notice of Professor Chodat and other botanists. Nevertheless, I think that the genus Golenkinia and the species G. radiata must give way to the genus Archerina and the species A. boltoni of nine years' earlier publication.

The name Phythelios given by Frenzel in 1891 to what is probably the same organism is also later than Archerina.

Whether there is anything like a constant or very frequent association of Archerina with a minute amœboid commensal remains an open question.

Subsequently to Chodat's description of Archerina as Golenkinia another botanist, Lemmermann, described (in 'Hedwigia,' Bd. xxxvii, 1898, p. 303) under the name "Richteriella botryoides," some of the phases of divi-

sion of Archerina, which were described and figured by me in my paper of 1885. These are the symmetrically-grouped division-products of Archerina drawn in figs. 21 and 22 of my paper (Pl. 7, 'Quart. Journ. Micr. Sci.,' vol. xxv). I would further draw attention to the oblong form of the chlorophyll bodies shown in my fig. 21, as agreeing with some of Lemmerman's figures of another supposed new genus of his.

It is, of course, possible to maintain that these oblong bodies are specially distinct from the more usual spherical forms, but I do not think that there is any sufficient ground for separating, generically or specifically, the much divided groups of small-sized spheres from the larger single spheres of Archerina boltoni with which they were associated and with which they are connected by intermediate phases of division, as well as by the characteristic radiating processes of the cell-envelope.

In my judgment Golenkinia and Richteriella are synonyms of Archerina, and I think this will be admitted by botanists who compare my plate of 1885 with the much later drawings of Chodat and of Lemmerman. I am at the same time of the opinion that the reference of Archerina to the Protozoa by me was an error, and that the organism is to be regarded as one of the simpler Protophyta.

B. Botryococcus.

Curiously enough it is to a paper also published by Prof. Chodat in Morot's Journal at a later date, 1896, that I am indebted for the identification of a very beautiful minute fresh-water organism which I studied and drew about the same time as that in which Archerina came before me. I used to speak of this as the "Cayenne pepper growth," since it appeared as little grains resembling in colour and size those of that condiment, floating in closely packed aggregates on the surface of the English Lakes (Grasmere and Derwentwater). I received it first in 1884 from Mr. Bolton, of Birmingham, and some sixteen years later from

my friend Prof. Hickson, F.R.S., of the University of Manchester. I was at first unable to identify it, but suspected it to be the Botryococcus of Kützing (1849), a suspicion which I was unable to confirm owing to the fact that no good figures of it were published. In 1896, however, Prof. Chodat published a coloured plate (Pl. III, p. 333) in Morot's 'Journal de Botanique' (which only came to my knowledge last year) accompanying a full account of the Botryococcus Braunii of Kützing, which he found abundantly and at various seasons of the year on certain parts of the surface of the Lake of Geneva. Prof. Chodat describes, and his figures illustrate, a purely green form of this organism, a phase which I have not seen. But he mentions that frequently the Botryococcus develops a brick-red coloured oil, which may be more or less abundant, and give a completely red appearance to the floating colonies. He points out that the red oily matter enables the organism to float, and expresses some doubt and interest as to the exact mode of formation of this red-coloured oil.

Whilst referring the reader to Prof. Chodat's memoir for many interesting observations, I will now briefly describe my own observations and the drawings made by me nearly twenty-five years ago, which I have never published, but now reproduce in Pl. 25 accompanying this paper.

General form and colour of the fronds.—The little "grains" of Botryococcus which float practically on the surface of the water in which it occurs are irregular, incomplete, hollow, spherical, or kidney-like bodies, connected one to another by growth and origin, and separating by rupture from one another after a certain size and shape has been attained. A group of these growths magnified about one hundred diameters is shown in outline in Pl. 25, fig. 4. In fig. 1 of Pl. 25 a smaller frond is shown more highly magnified. This drawing also serves to show the very striking coloration of the first specimens which came under my notice, viz. a golden-red mass with a translucent green-coloured cortex. The colouring is so strong as to recall the

beautiful tints of uranium-glass and to suggest the adjective "chryso-chlorous" to describe this green and gold phase of vegetation. A more highly magnified view of a portion of the edge of the frond as seen when under the pressure of a cover-glass (Pl. 25, fig. 3) shows that the two colours are due (*a*) to the green colour of the granules, and to some extent of the cell-substance of the cell-units which build up the organism and form its superficial layer; and (*b*) to the orange-yellow or often brick-red colour of the jelly which is formed by the cell-units and holds them together.

As shown in fig. 2 and fig. 3 of Pl. 25 droplets of a more or less oily nature are pressed out of the jelly when a cover glass is laid over it.

In many specimens I found the cells pale green with green granules whilst the jelly was brick-red. In other cases the cells were as just stated, but the jelly was yellow. I did not find any instances in which the jelly was green in colour, though Prof. Chodat has figured the fronds in that condition.

In some examples I found that, whilst the general substance of the cell had a sage-green tint, the granules were orange-yellow. These are shown in Pl. 25, figs. 6 to 10. In some specimens, which were of very strong brick-red colour, the cells contained a very large quantity of orange-red granules (Pl. 25, figs. 11, 12, and 13).

I do not gather from Prof. Chodat's account of Botryococcus that it has ever been shown that the green colouring matter is chlorophyll. My own impression, based chiefly on the sage-green tint, was that it was not. But the nature of the green pigment seems not to have been definitely determined. I mention this because in an organism which has very much smaller cell-units, but forms similar hollow botryoidal colonies, namely, Clathrocystis æruginosa, Henf., the green colour, though the alga is of an intense apple-green when collected in mass (as a sort of cream) is certainly not due to chlorophyll, but to a peculiar body insoluble in alcohol and changed by ether into a brown pigment. The same

peculiar green pigment occurs in Aphanizomenon flos-aquæ, with which I believe Clathrocystis to have a special relationship. This pigment is remarkable for changing, when dried with exposure to light and air, from an apple-green to a blue verdigris-green tint.

The jelly.—The cell-units of Botryococcus form a closely-set superficial layer one cell deep. They secrete a jelly-like material which forms a denser capsule to each cell (Pl. 25, figs. 16 and 18), and is of a softer more watery consistence below that layer and between the adjacent capsules. Under slight pressure the capsules burst, and the cell itself is shot out of its position in the jelly into the surrounding water. The capsules burst by dehiscence of a concave-convex lid (fig. 18).

In the parts where one sub-spherical or kidney-shaped mass is adherent to similar neighbouring masses the jelly is often broken up into fibrillated strands (Pl. 25, fig. 5), the formation of which seems to be connected with the division of one original colony into separating sub-colonies.

The colour of the jelly in all my specimens was either golden-yellow or a deeper brick-red. I gather from Prof. Chodat's description that it may present itself as entirely colourless or with a greenish tint.

I made no chemical tests of the nature of this jelly, but some are recorded by Chodat.

The cell-units.—These as shown in the figures in Pl. 25 are oblong and somewhat pyriform. They consist of a continuous dense substance, which rarely exhibits vacuoles (fig. 17). From one to forty sharply marked granules are embedded in the dense substance and these granules are in some specimens green (fig. 3), in others they are yellow (figs. 9, 10), in others brick-red (figs. 11, 12, 13).

The cells of Botryococcus when extruded from their capsules and the adjacent jelly exhibit no movement. They are devoid of flagellum or cilia.

In the living cells it is not possible to observe any structure in the cell representing a nucleus, but I found in

specimens stained with hæmatoxylin en masse for several hours that a central substance exists which shows a deep purple stain (see Pl. 25, fig. 14). I did not detect any characteristic nuclear structure in this in specimens clarified and mounted in the usual way, but I am not in a position to say that such might not have been present though it escaped my observation.

The cells appear to divide by binary fission along the long axis (figs. 15, 17). After fission the resulting cell-units separate and recede to a distance from one another equal to the short diameter of the cell, the interspace being filled by jelly (fig. 18).

It is a long time since I had the opportunity of observing this organism, and I should urge those who may meet with it in the Lake district or elsewhere to direct their attention to the following points:

(*a*) The nature of the green colouring matter.

(*b*) The relation of the variable amount of yellow and red oily pigment to the season.

(*c*) The mode of passage of the colouring matter into the jelly.

(*d*) The existence of specimens showing colourless and of others showing green-coloured jelly.

(*e*) The nuclear structure.

(*f*) The possible occurrence of other modes of reproduction than the longitudinal fission leading to increase in the size of colonies.

It does not appear that more than one European species of Botryococcus can be distinguished. The genus Ineffigiata of Mr. West is, I am informed by him, probably based upon a local growth variety of Botryococcus Braunii.

May 31*st*, 1908.

EXPLANATION OF PLATE 25,

Illustrating Sir Ray Lankester's Memoir "On Archerina, Golenkinia, and Botryococcus."

Fig. 1.—A floating colony of Botryococcus Braunii, Kützing, of the chrysochlorous variety. Magnified about 100 diameters.

Fig. 2.—Portion of a colony under cover-glass pressure, superficial focus. More highly magnified.

Fig. 3.—A portion of a reddish colony. Still more highly magnified.

Fig. 4.—Outline of a series of contiguous hollow reniform colonies, as found floating on lake-water. Magnified about 50 diameters.

Fig. 5.—Portion of neighbouring reniform masses more highly magnified to show the fibrillated strands of jelly connecting them to one another.

Fig. 6.—Cell in the gelatinous investment, with yellow granules.

Fig. 7.—Cell with displaced capsule-lid.

Figs. 8, 9, 10.—Similar cells to that shown in Fig. 6.

Figs. 11, 12, 13.—Large cell units with abundant orange-coloured granules.

Fig. 14.—Five cells treated with alcohol, followed by hæmatoxylin and alum-staining-fluid, and mounted in balsam. A dark stained central body is seen.

Fig. 15.—Cells in binary longitudinal fission. Observe the vacuoles.

Fig. 16.—Cell with strongly marked capsule.

Fig. 17.—Cells in binary longitudinal fission, as in Fig. 15.

Fig. 18.—Two adjacent cells to show the shell-like capsule of each, the interposed jelly, and the capsular lid of each cell reflected owing to pressure, and about to allow the cell to be ejected as a naked isolated unit.